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Pneumocystis carinii, Toxoplasma gondii, Cytomegalovirus and the Compromised Host

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Pneumocystis carinii and Toxoplasma gondii are the two major parasitic protozoan pathogens in the immunocompromised host. Both organisms cause latent infection in humans and many animals. Cats are the definitive hosts for toxoplasmosis; the animal vector for pneumocystis (if any) has not been defined. Toxoplasma is an obligate intracellular parasite, whereas the available evidence suggests that Pneumocystis carinii exists primarily extracellularly. In compromised hosts, pneumocystis infection usually involves only lungs, whereas toxoplasma causes a generalized infection with encephalitis being the principal clinical manifestation. Both types of infection are treated with combinations of folate antagonists (trimethoprim or pyrimethamine with sulfonamide). Both parasites are associated with cytomegalovirus infection in immunosuppressed hosts, an association which may be due to symbiosis between parasites, or to an additive immunosuppressive effect of dual infection on the hosts.

IT IS COMMON knowledge that infection is a frequent cause of morbidity and mortality in patients with defective host defenses. Pneumocystis carinii and Toxoplasma gondii are the only protozoa that frequently cause disease in the compromised host. Resistance to infection with these parasites is generally thought to be due to cell-mediated immunity (that is, thymus-derived-lymphocytes [T-lymphocytes] and macrophages), although there is evidence that humoral immunity plays some role in host resistance to infection by both organisms.

Although Pneumocystis and Toxoplasma will be the major focus of this discussion, infection with these two organisms cannot be discussed without reference to cytomegalovirus. This agent is isolated frequently from immunocompromised patients, although many infections are of minimal clinical significance. In addition, dual virus-parasite infections occur frequently, perhaps more commonly than would be expected by chance. At the conclusion of this report, we will examine some of the evidence that infection with one of these organisms, especially cytomegalovirus, may facilitate secondary infection by Pneumocystis carinii or Toxoplasma gondii.

In recent years, the obligate intracellular protozoan Toxoplasma gondii has emerged as a significant pathogen of man and animals. It is one of the most common infections of man. In the United States, approximately 50 percent of the population harbor the latent form of the organism.[1] Most infections in the normal host are asymptomatic, and clinically apparent infections are usually self-limited.[2] In pronounced contrast, toxoplasmosis in the compromised host is frequently fatal.[1]

History

Toxoplasma was discovered in 1908 when Nicolle and Manceaux[3] observed a parasite in

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ABBREVIATIONS USED IN TEXT
IFA = immunofluorescent antibody
SMX = sulfamethoxazole
TMP = trimethoprim

mononuclear cells of a North African rodent, the gondi. The first recognized case of toxoplasmosis in humans was described by Jankû in 1923.[4] However, it was not until 1937 that toxoplasmosis as a disease in humans emerged as a potentially serious problem when Wolf and Cowen[5] described a case of congenital infection in a neonate and went on to establish Toxoplasma as a cause of prenatally transmitted human disease. In 1940 Toxoplasma was recognized as a cause of disease in adults when Pinkerton and Weinman[6] described a fatal case in a young man. In 1948 Sabin and Feldman[7] developed a serologic test for the detection of Toxoplasma antibody that greatly facilitated investigation of the epidemiologic and clinical aspects of toxoplasmosis. Although reports describing some of the clinical manifestations of Toxoplasma infection in the normal host appeared first in the 1950's,[8,9] it was not until 1968 that Vietzke and his colleagues[10] at the National Cancer Institute published the first comprehensive study of immunosuppressed patients with toxoplasmosis. These investigators noted a predilection of this organism for the central nervous system and they described the frequent occurrence of concomitant disseminated infection with cytomegalovirus. Finally, it was not until 1969—more than 60 years after the discovery of the parasite—that the cat family was identified as the definitive host of Toxoplasma.[11]

The Organism

Toxoplasma exists in three forms: the trophozoite, the tissue cyst and the oocyst (Figure 1).

The trophozoite (Figures 1A, 1B) of T. gondii is crescentic or oval, approximately 2 μm to 4 μm in width, and 4 μm to 8 μm in length. It is the rapidly proliferating form of the organism seen in tissues during acute infection. The trophozoite can enter all mammalian cells except possibly the nonnucleated erythrocyte.[12] It is easily killed by freezing and thawing, by desiccation and by gastric and duodenal digestive juices.[13] Because it is an obligate intracellular parasite, it does not grow on artificial media but may be propagated in laboratory animals, embryonated eggs and cultured or mammalian cells.

Like most obligate intracellular parasites, T. gondii enters cells by a process called endocytosis whereby the organism is enveloped by host cell pseudopodia that fuse and enclose the parasite within a cytoplasmic vacuole.[14] In the case of cells generally not considered phagocytic, there is controversy concerning the role of the host cell in the entry process.[15] Several investigators have data that they interpret as showing that Toxoplasma can induce phagocytosis in cells such as the fibroblast,[16,17] whereas others have data supporting the view that the parasite is the only active agent in the entry process.[15,18] The controversy remains unresolved.

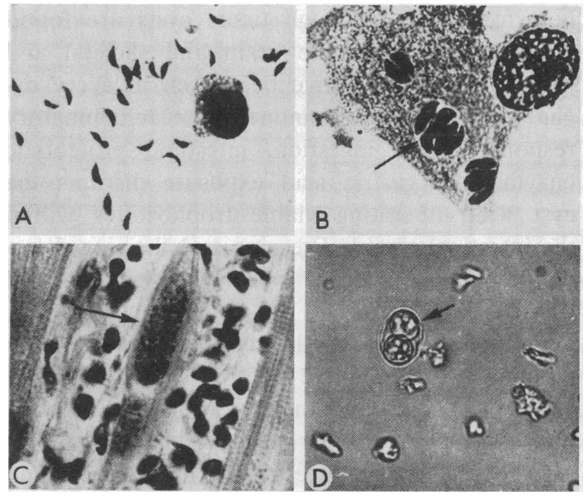

Figure 1.—The three forms of Toxoplasma. **A,** Trophozoites from peritoneal fluid of an infected mouse. **B,** Trophozoites in cytoplasmic vacuoles of human macrophage cultured *in vitro*. Notice that the parasites have replicated, forming a pair, tetrad and rosette **(arrow)**. **C,** Tissue cyst **(arrow)** in human myocardium. **D,** Oocyst **(arrow)**. (Reprinted by permission from Anderson S, Remington JS: South Med J 68:1433-1443, Nov 1975.[40])

After cell entry, Toxoplasma undergoes a resting phase and then begins to multiply in cytoplasmic vacuoles, eventually rupturing the cell. Toxoplasma overcomes the microbicidal mechanism of phagocytes, such as macrophages, by preventing lysosomes from fusing with phagosomes in which the parasite resides.[16] The mechanism by which it accomplishes this is not known, but dead Toxoplasma or organisms coated by specific Toxoplasma antibody do not prevent phagolysosome formation and are degraded.[16]

The tissue cyst (Figure 1C) is the latent form of the organism. It is formed by trophozoites within a host cell vacuole. The factors that trigger tissue cyst formation are unknown, but they may be related in part to the development of immunity. Cysts containing viable trophozoites persist in the host for life.[19] Although tissue cysts are found most commonly in the brain and skeletal and heart muscle, they have been seen in every organ.[19] Cysts evoke minimal host inflammatory response. After ingestion, intestinal proteolytic enzyme and gastric acid exposure disrupts the cyst wall, liberating viable trophozoites within the intestinal tract.[13] Freezing and thawing, heating above 60°C (140°F) and desiccation will destroy tissue cysts.[13]

Oocysts are produced only by members of the cat family, the definitive host of T. gondii (Figure 1D). Oocyst production takes place within the intestinal epithelium. A cat may shed as many as 10^7 oocysts in the feces in a single day,[20] and production of oocysts continues for an average of 14 days.[20] Excreted cysts become infectious only after sporulation, which takes from a day to 21 days depending on environmental factors.[21] As immunity to the intestinal epithelial form of toxoplasmosis in the cat is not complete, production of oocysts can recur after reinfection.[22] The oocyst is the most hardy form of T. gondii; under optimal conditions, it may survive up to a year in soil.

Epidemiology

Worldwide in distribution, toxoplasmosis is one of the most common infections of man. The prevalence of Toxoplasma antibody increases with age, and there is no difference in the prevalence between sexes.[23] Infection occurs naturally in all mammals, some birds, and probably some reptiles.[24] Prevalence of infection varies considerably with geographic location (Table 1). The prevalence of Toxoplasma antibody is higher in the warmer and moister areas of the world than in colder regions.[23] In addition, antibody is more prevalent in populations living at low altitudes than at high altitudes.[25] Toxoplasma infection may be transmitted prenatally or postnatally. Congenital transmission can occur only during infection of a pregnant woman lacking antibody to Toxoplasma.[26] The likelihood of transmission of infection from mother to fetus increases with each trimester of pregnancy; however, clinically apparent disease in the neonate occurs most commonly when the fetus is infected in the first trimester.[27]

TABLE 1.—*Prevalence of Antibodies to Toxoplasma in Different Population Groups**

Population	*Positive Percent*
Iceland	11
Portland, Oregon	17
St. Louis	27
New Orleans	31
Pittsburgh	35
Haiti	36
Honduras	64
Tahiti	68

*Adapted from Feldman and Miller.[23]

Infection acquired after birth is most likely to occur by the oral route. Ingestion of raw or undercooked meat containing tissue cysts is one source of infection; the commercially available meats most likely to contain tissue cysts are lamb and pork.[13] Infection by the oral route also may occur if material is ingested that is contaminated with oocysts from cat feces. The relative role of each of these sources in the overall prevalence of toxoplasmosis in man has not been determined. Several other sources of Toxoplasma infection in humans have been documented (Figure 2). Toxoplasma has been transmitted to seronegative re-

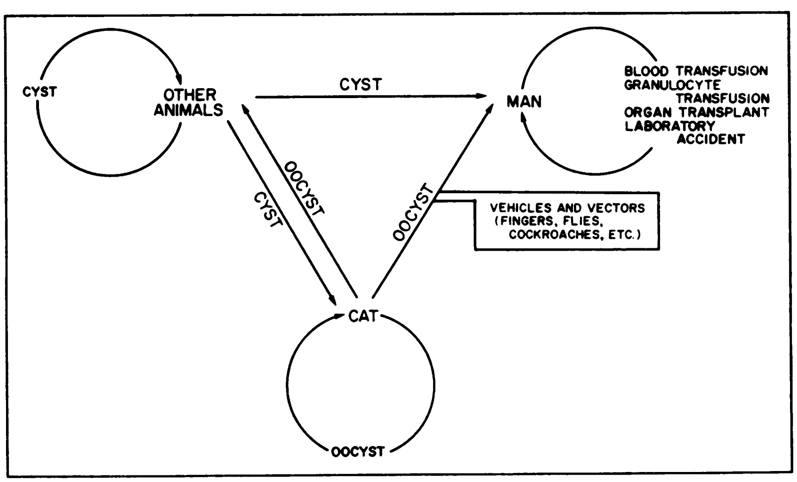

Figure 2.—Transmission of Toxoplasma gondii. (Reprinted by permission from Krick JA, Remington JS: N Engl J Med 298:550-553, Mar 1978.[1])

cipients by transfusion of blood products. In a retrospective study, Siegal and associates[28] documented that transfusion of granulocytes from donors with chronic myelogenous leukemia transmitted Toxoplasma infection to four patients with acute leukemia. Probable transmission of toxoplasmosis by means of organ transplantation has recently been observed (Ryning and co-workers, unpublished observations). In this study, two heart transplant recipients who had no serologic evidence of latent or active Toxoplasma infection at the time of operation developed clinically apparent toxoplasmosis after surgery; one died of disseminated toxoplasmosis and the other survived but had significant neurologic sequelae. Because both heart donors had evidence of recently acquired toxoplasmosis at the time of operation, the transplanted hearts represented the most likely source of infection. Finally, transmission of Toxoplasma infection as a result of laboratory accidents has been described.[29]

Pathogenesis

The walls of ingested oocysts or tissue cysts are degraded by gastric acid and intestinal enzymes. The resulting liberated organisms invade the intestinal epithelium, undergo intracellular multiplication and subsequently disseminate by means of the blood stream to virtually every organ and tissue. Toxoplasma trophozoites invade and destroy host cells, resulting in foci of tissue necrosis that evoke a predominantly mononuclear inflammatory cell response. The host response to infection includes production of antibodies as well as development of cell-mediated immunity. Because intracellular organisms are protected from the lethal effect of antibody and complement,[16] cell-mediated immunity appears to be crucial for the control of Toxoplasma infection as clearly demonstrated in animal experiments.[30] The macrophage is an important component of the effector arm of cell-mediated immunity. Because Toxoplasma can multiply in normal macrophages, effective control of Toxoplasma infection through this arm of the immune response mandates a functional alteration of the macrophage whereby it acquires the ability to kill or inhibit the multiplication of Toxoplasma, a process called macrophage activation. Activation occurs when T-lymphocytes sensitized to Toxoplasma antigen elaborate soluble products called lymphokines in response to Toxoplasma antigen. These lymphokines alter macrophage morphology and function, resulting in activation. The biochemical basis of lymphokine activation of macrophages is largely unknown.

TABLE 2.—*Underlying Conditions Associated With Toxoplasmosis in the Compromised Host**

Underlying Condition	*Percent of Cases*
Malignancies	
Hodgkin disease	40
Non-Hodgkin lymphoma	11
Leukemia	19
Solid	12
Other	2
Collagen-vascular disease	7
Organ transplant	9

*Adapted from Ruskin and Remington.[37]

Clinical Manifestations

Toxoplasmosis may be classified into four categories: (1) congenital, (2) acquired toxoplasmosis in the normal host, (3) acquired or reactivated toxoplasmosis in the immunocompromised host and (4) ocular toxoplasmosis. This discussion will be limited to Toxoplasma in normal and compromised hosts.

Normal Host

Most Toxoplasma infections in the normal host are asymptomatic. Clinical manifestations are usually mild and self-limited, with spontaneous resolution occurring in most cases within one to three months. The most common clinical manifestation is asymptomatic localized lymphadenopathy,[2] which most frequently involves the cervical nodes. Generalized adenopathy can occur. Fluctuation in lymph node size is common and persistence of lymphadenopathy for more than a year has been reported.[31] In addition to lymphadenopathy, various other signs and symptoms such as fever, malaise, pharyngitis and splenomegaly may occur. At times toxoplasmosis may be clinically indistinguishable from infectious mononucleosis caused by Epstein-Barr virus infection. The presence of atypical lymphocytes can add to the confusion.[2] A skin rash may occur; it is generalized, usually nonpruritic, transient and most pronounced on the trunk and proximal extremities.[31] Illness may persist for months, and fluctuations in signs and symptoms are not uncommon.

Clinical illness reflecting isolated organ involvement in the normal host has been described but is uncommon. Myocarditis secondary to destruction of myocardial cells by trophozoites may cause serious consequences.[31] Pericarditis may occur but is rare.[32] Toxoplasma has been implicated as a cause of pneumonia,[33] myositis,[34] and hepatitis with a clinical course and liver enzyme rise similar to that of viral hepatitis.[35] Toxoplasma-induced encephalitis in the normal host is rare. Chorioretinitis due to Toxoplasma has been observed in the normal host during the course of acquired infection, but most cases probably represent activation of congenitally acquired disease.[36]

Compromised Host

Toxoplasma gondii has emerged as an important opportunistic pathogen in the compromised host that is capable of producing rapidly fatal disseminated infection. Sources of infection in this setting include reactivation of latent infection during immunosuppression and introduction of organisms into a host with impaired immunologic mechanisms.

Toxoplasmosis in the compromised host occurs most frequently in patients with lymphoreticular malignancies, particularly Hodgkin disease, but it has been associated with a wide variety of underlying conditions including hematologic malignancies, solid tumors, collagen-vascular disease and organ transplantation (Table 2).[37] It is unusual for toxoplasmosis to occur unless the patient is receiving immunosuppressive therapy.[37]

In the compromised host, the central nervous system is the most common site of involvement. There is evidence of meningoencephalitis in more than 90 percent of immunosuppressed patients with toxoplasmosis.[1] The neurologic signs and symptoms are nonspecific (Table 3), reflecting diffuse or focal involvement. The most common manifestation is alteration of mental status. Mass lesions may occur, mimicking a brain abscess.[38] The cerebrospinal fluid abnormalities also are nonspecific. Typical findings include mild pleocytosis with a predominance of mononuclear cells, a normal glucose level and mild-to-moderate elevations in protein concentration. Although Toxoplasma has a predilection for the central nervous system, virtually any organ system can be involved. Necrotizing myocarditis and interstitial pneumonia are not uncommon.[1] Manifestations of Toxoplasma infection such as lymphadenopathy or hepatosplenomegaly that are common in the normal host also may occur in the immunocompromised patient. Persistent fever, unresponsive to antibiotics, may be the initial manifestation of Toxoplasma infection.[39] Unfortunately, the manifestations of Toxoplasma infection in the compromised host are nonspecific and similar to those that may be produced by the patient's underlying disease or other opportunistic pathogens.

TABLE 3.—*Neurologic Signs and Symptoms of Toxoplasmosis in the Compromised Host**

Signs or Symptoms	*Percent*
Alteration of mental status	68
Motor impairment	32
Seizures	24
Abnormal reflexes	16
Headache	14
Vomiting (projectile)	5
Sensory impairment	5

*Adapted from Remington.[2]

Diagnosis

The diagnosis of acute toxoplasmosis can be made by (1) serologic tests, (2) histologic examination of infected tissue and (3) isolation of the parasite.

Serologic Testing

Serologic tests for the detection of Toxoplasma antibody constitute the primary mechanism of diagnosis of acute and chronic infection. The humoral response to Toxoplasma infection usually includes production of both IgG and IgM antibodies. IgM antibodies are detected using the immunofluorescent antibody (IFA) technique. The usefulness of this test is based on the observation that IgM antibodies appear before IgG antibodies and persist for only a relatively brief period in most patients. The test was originally designed to facilitate the diagnosis of congenital infection by differentiating passively transferred antibody (IgG) from the infant's own IgM antibody response. However, it is also quite useful in the diagnosis of acute infection in the normal and compromised host.

IgM antibodies usually appear during the first week of infection, rise rapidly, peak within the first month, and then decline and disappear three or four months after onset of the infection. In some cases IgM antibody may disappear as early as three weeks after infection.[1] During acute infection, IgM Toxoplasma titers can vary widely from 1:10 to greater than 1:1,000, but only a fourfold or greater rise or an extremely high titer (1:160 or higher) establishes the presence of acute infection.[40] A low titer does not exclude the possibility of active infection.[40] Although in most cases IgM antibodies disappear within three to four months, in some cases IgM antibodies have persisted for months or even years but usually at low titers of 1:20 or less.

IgG antibodies may be detected by IFA (the test most commonly used), the Sabin-Feldman dye test, indirect hemagglutination, or complement fixation. As with the IgM-IFA test, a fourfold rise in IgG antibody titers between serum samples run in parallel establishes the diagnosis of acute infection.[40] Titers measured by these tests rise more slowly than IgM titers, at times taking two months or more to peak.[1] In most cases, the maximum titer usually exceeds 1:1,000, and titers of 1:10,000 and higher have been reported.[1] Because high titers of IgG antibody have been known to persist for years, an isolated high titer does not establish the presence of acute infection.[1] After four to six months, the IgG titer slowly begins to fall, but usually some antibody remains indefinitely.[40] Treatment may blunt maximal antibody response if it is begun early enough; however, once maximal levels are reached, treatment rarely has an effect.[27] Complement-fixing and hemagglutinating antibodies usually develop later and rise more slowly than IgG-IFA or dye test antibodies.[40]

Histologic Diagnosis

A definitive diagnosis of acute toxoplasmosis can be made by the demonstration of trophozoites in tissues or body fluids.[40] The recognition of trophozoites in histologic sections is difficult but can be facilitated by using Giemsa or Wright stain rather than hematoxylin and eosin. Immunofluorescent staining also may facilitate identification of organisms.[41] The presence of cysts in tissue usually represents subacute or chronic infections; but, because these structures can form early, their presence does not exclude the possibility of acute infection. Lymph node architecture during Toxoplasma lymphadenitis is sufficiently characteristic to be considered diagnostic.[42]

Isolation

Isolation of the organism from body fluids (for example, cerebrospinal fluid or aqueous humor) establishes the diagnosis of acute Toxoplasma infection. This is usually accomplished by injecting suspect fluids into the peritoneal cavity of a mouse. Use of this method frequently delays establishing the diagnosis. Because the organisms can be recovered from tissues containing cysts, their isolation from this source does not necessarily indicate acute infection.

Summary—Diagnosis

Normal Host

The methods most commonly employed in the diagnosis of acute Toxoplasma infection in the normal host are summarized in Table 4. The demonstration of a fourfold rise of IgM or IgG antibodies or the presence of an extremely high IgM titer (excluding false-positive results due to the presence of rheumatoid factor and antinuclear antibody) establishes the diagnosis of acute infection. Histologic examination of a lymph node

TABLE 4.—*Guide to the Diagnosis of Toxoplasmosis**

Syndrome	*Definitive Diagnosis*	*Presumptive Diagnosis*
Acute acquired toxoplasmosis in the immunocompetent host	1. Rising antibody titer 2. Markedly elevated titers of IgM-IFA ($\geq$1:160) in presence or absence of symptoms 3. Characteristic histology on lymph node biopsy	1. Elevated titer of DT or IFA ($\geq$1:1,000) and positive IgM-IFA test associated with characteristic clinical syndrome (for example, lymphadenopathy, "mononucleosis-like" syndrome)
Acute toxoplasmosis in the immunodeficient host	1. Rising antibody titer 2. Markedly elevated titers of IgM-IFA ($\geq$1:160) 3. DT titer per IgG concentration greater in cerebrospinal fluid than in serum 4. Histologic or cytologic demonstration of trophozoite (for example, brain or lung biopsy, bone marrow aspirate, cerebrospinal fluid sediment) 5. Isolation of parasite from cerebrospinal fluid	1. Elevated titer of DT, IFA, CF, IHA, or IgM-IFA associated with characteristic clinical syndrome (for example, encephalitis, pneumonitis, myocarditis, lymphadenopathy)

CF = complement fixing antibody
DT = Sabin-Feldman dye test antibody
IFA = immunofluorescent antibody
IHA = indirect hemagglutinating antibody

*Adapted from Anderson and Remington.[40]

also can secure the diagnosis. A presumptive diagnosis of acute Toxoplasma infection may be made in the presence of an elevated IgG titer (1:1,000 or higher) and a positive IgM-IFA test associated with characteristic clinical manifestations.

Compromised Host

The criteria for the definitive and presumptive diagnosis of acute toxoplasmosis in the compromised host also are summarized in Table 4. The same serologic criteria for the definitive diagnosis of acute toxoplasmosis in the normal host apply to the compromised host. False-negative serologic tests may occur.[1] Although not frequently used, the demonstration of local central nervous system antibody production by finding a greater Toxoplasma IgG titer per gram of globulin in cerebrospinal fluid than in the serum is diagnostic of active infection. Finally, isolation of trophozoites from body fluids also is definitive proof of infection.

Treatment

Sulfadiazine or triple sulfonamides combined with pyrimethamine is the standard treatment regimen for toxoplasmosis. Both drugs are used because the combination is synergistic against Toxoplasma.[43] Therapy in the normal host is usually not necessary because of the self-limited nature of the infection but is indicated when vital organs are infected or severe constitutional symptoms persist.[1]

In the compromised host, therapy is always indicated because most patients will die if they are not treated.[37] Retrospective studies have shown an 80 percent response rate if therapy with sulfadiazine and pyrimethamine is instituted early in the course of the illness.[37] Toxicity can occur with these drugs. The side effects of the sulfonamides are well known; pyrimethamine may cause bone marrow suppression, but the risk can be minimized by the administration of folinic acid and fresh yeast.

Prevention

An important aspect of the control of Toxoplasma infection is the prevention of infection in the compromised host. Meat should be stored at −20°C (−4°F) or heated to 60°C (140°F) for 15 minutes to kill tissue cysts. Hands should be washed before eating and after touching uncooked meat. Cat feces or material potentially contaminated by cat feces should be avoided. Periodic serologic testing is extremely important to facilitate the diagnosis of Toxoplasma infection. Because this organism can be transmitted by means of blood, blood products or transplanted organs, it may be prudent to use seronegative donors.

Dual Infections

There has been ample documentation of concomitant infection with Toxoplasma and cytomegalovirus in the compromised host.[44,45] In 1956 Hemsath and Pinkerton[46] described simultaneous disseminated cytomegalic inclusion disease and toxoplasmosis in an adult with myeloid metaplasia. In 1968 Vietzke and colleagues[10] at the National Cancer Institute were the first to call attention to the striking concurrence of disseminated cytomegalovirus and Toxoplasma infection in patients with altered host defenses. They suggested that this association might be more than a fortuitous occurrence and that there might be a symbiotic relationship. Individual human cells infected with both organisms also have been observed.[10] This association is discussed more fully at the end of this report.

Pneumocystis Carinii

Pneumocystis carinii is an organism that has not been classified definitely, although it is commonly believed to be a protozoan. In 1909 Chagas[47] in Brazil first observed the organism in the lungs of guinea pigs experimentally infected with Trypanosoma cruzi. He later saw similar parasites in the lungs of a man dying of American trypanosomiasis.[48] Carini, also working in Brazil, found similar organisms in the lungs of rats experimentally infected with trypanosomes; both investigators thought they were specialized forms of trypanosomes.[49] In 1912 Madame and Monsieur Delanöe[50] reviewed Carini's slides and realized that they had observed this organism in the lungs of Parisian sewer rats not infected with other protozoans, thereby distinguishing it as a separate parasite. They named the organism Pneumocystis carinii to indicate its apparent tissue tropism and to honor one of its discoverers. At that time, no animal or human disease was attributed to this parasite.

In the early years of World War II, two German physicians, Ammich[51] and Beneke,[52] described a nonsyphilitic interstitial plasma cell

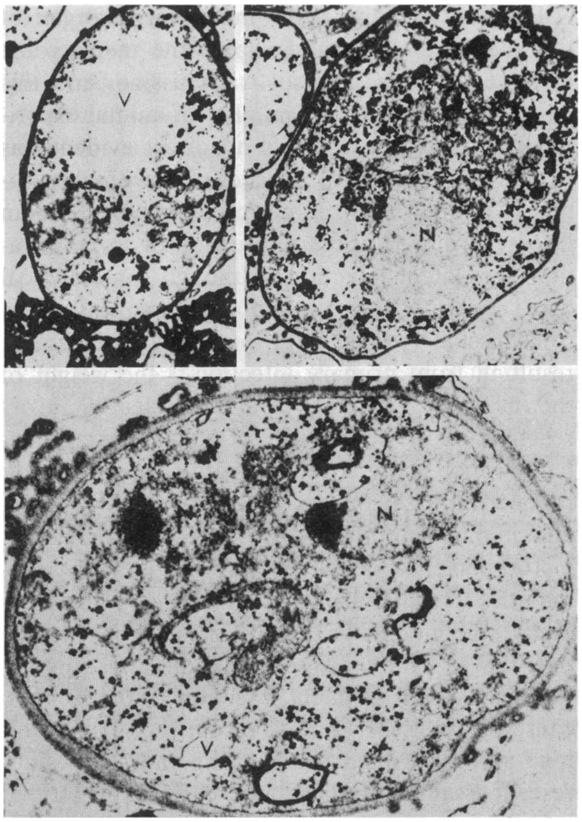

Figure 3.—Electron micrographs of Pneumocystis carinii in infected rat lung, stained with lead citrate. **Top:** Trophozoites with early thickening of surface pellicle, vacuole (V), nucleus (N) and numerous mitochondria (×15,000-21,000). **Bottom:** Cyst with fully-developed trilaminar wall. Nuclei (N), mitochondria (M) and vacuoles (V) can be seen (×37,000). (Reprinted by permission from Barton EG Jr, Campbell WG Jr: Am J Pathol 54:209-236, Feb 1969.[61])

pneumonia in human neonates and infants. This illness assumed epidemic proportions in Europe during the years of World War II, especially among malnourished infants in foundling homes.[53] Although Ammich, Beneke and subsequent investigators during the war years were not able to implicate an etiologic agent in this illness, review of their published photomicrographs has shown the presence of P. carinii. In 1942 two Dutch investigators, Van de Meer and Brug,[54] found P. carinii in the lungs of three patients post mortem, although there was no associated clinical illness or inflammatory response. It was not until 1951 that a Czechoslovakian worker, Vanêk,[55,56] solidly associated P. carinii infection with plasma cell pneumonia of children. His work was widely discounted in the United States, however, and it was not until several years later that the Nobel laureate D. Carleton Gajdusek lent his influence to this observation,[57] and P. carinii became accepted as the principal etiologic agent of interstitial plasma cell pneumonia in infants and children. In 1955, Hutchison[58] reported an association between P. carinii pneumonia and congenital immunodeficiency. Pentamidine isethionate was first used for treatment in 1963,[59] but related aromatic diamidines were used as early as 1957.[60]

The Organism

What sort of organism is P. carinii? Because it has only recently been grown *in vitro,* all of the evidence to date that we have about its structure and life cycle derives from morphologic observations on organisms grown in animal lungs. Barton and Campbell[61] have published the definitive work on the electron microscopic morphology of this organism in rats, and similar observations have been made on material from human lungs.[62] Two forms can be distinguished, the trophozoite and the cyst, with intermediate forms suggesting that trophozoites develop into cysts containing four to eight trophozoites. Fully developed cysts apparently rupture and release trophozoites into the surrounding environment. Based on electron microscopic observations of infected rat lungs, Vossen and co-workers[63] suggested that partially developed cysts also may release trophozoites. Figure 3, from Barton and Campbell's studies, shows trophozoite forms in the two top panels. They contain intracytoplasmic organelles typical of eukaryotic cells, such as a nucleus, endoplasmic reticulum and mitochondria. An early cyst form is shown in the lower panel, and in Figure 4 a mature cyst form is shown, with four trophozoites contained within it. Morphologically, the trophozoites resemble those of T. gondii, except that Pneumocystis trophozoites lack specialized polar structures thought to be used for cell entry.

The available evidence strongly supports inclusion of P. carinii in the phylum Protozoa, probably in the class Sporozoa.[64] In experimental animals, infection with this organism responds to antiprotozoal drugs such as the aromatic diamadines and folate antagonists and does not respond to antifungal agents such as amphotericin B or to antibiotics.[65] The organism does not grow on acellular media that support the growth of most fungi and bacteria,[66] and it has a broad host range similar to that of Toxoplasma.[64] The electron microscope shows intracellular organelles similar to those found in eukaryotic cells and not found

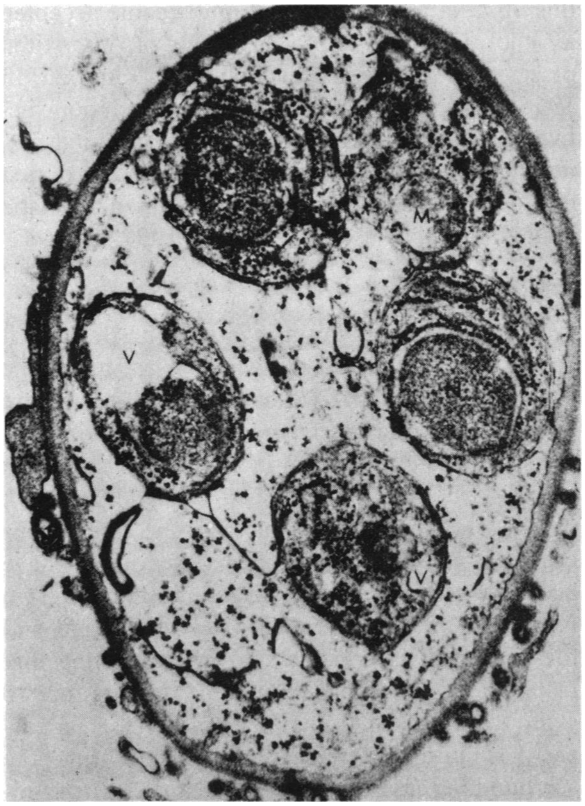

Figure 4.—Electron micrograph of fully developed cyst of Pneumocystis carinii from infected rat lung, stained with lead citrate. Five intracystic bodies (? immature trophozoites) are seen. Mitochondria (M), nuclei (N) and cytoplasmic vacuoles (V) are seen (×50,000). (Reprinted by permission from Barton EG Jr, Campbell WG Jr: Am J Pathol 54:209-236, Feb 1969.[61])

in prokaryocytes such as fungi and bacteria.[61-63] Now that the organism has been grown *in vitro*[66-68] it seems likely that metabolic studies will be done to precisely define its taxonomic status.

Epidemiology

Pneumocystis has been found in all geographic locations where an effort has been made to find it.[69] It has an extraordinarily wide host range: it has been demonstrated in the lungs of many higher primates; a number of domestic animals, including horses, sheep, goats, pigs, dogs and cats; some wild animals, such as foxes, and a number of rodent species, including rats, guinea pigs, mice and rabbits.[64] Although most of these observations have been incidental ones, without associated disease in the host, there are a few reports of clinically significant Pneumocystis pneumonia in animals. For example, a recent case report in the *Journal of the American Veterinary Association* describes two cases of fatal Pneumocystis pneumonia in Arabian colts.[70]

Because the lung is the major target organ of this parasite, one would expect the major route of infection to be aerosol transmission, and this has been confirmed in a number of animal experiments.[71] However, there is no direct evidence as to the mechanism of spread to or among humans. In addition, it is not clear whether human disease is due to primary infection or to reactivation of a latent infection. Although some clinical case reports suggest possible human-to-human transmission (primarily nosocomial) with disease resulting from primary infection,[72] the weight of the evidence presently available clearly favors reactivation in the majority of patients with pneumocystosis. Many strains of rats (and also other rodents) are asymptomatic carriers of P. carinii, and immunosuppression in these animals results in 100 percent mortality from Pneumocystis pneumonitis.[65] In humans, the incidence of Pneumocystis pneumonitis is directly proportional to the intensity of immunosuppressive therapy, suggesting reactivation of latent infection as the primary cause of disease.[73] Finally, recent data from Holland and the United States, using an immunofluorescent assay for antibody to P. carinii, suggest that infection is widespread and occurs early in life (Figure 5).[74-77] By the age of four years, the prevalence of antibody to P. carinii is 75 percent to 80 percent, and it remains at this level throughout life. These data suggest that, as in the other animal species studied so far, human infection with Pneumocystis is widespread and that disease results from reactivation of latent infection when host defenses are compromised by immunosuppressive agents or other factors.

Predisposing Conditions

What are the clinical conditions that are associated with P. carinii pneumonitis in humans (Table 5)? Historically, it has been associated with prematurity and starvation or malnutrition in children.[53,57,59] The disease was described subsequently in patients with congenital immunodeficiency of either the humoral or cellular type.[78] More recently, it has been reported in adults with acquired immunodeficiency, either due to disease or drug therapy.[79,80] Because there are data now to support the notion that both prematurity and starvation are associated with defects in cell-mediated and humoral immunity,[81] we may assume that the common denominator of P. carinii infection is immunodeficiency. Although most of the conditions associated with clinical pneumocytosis appear to be defects predominantly of cell-mediated immune function, there are well-described cases in children with isolated humoral immunodeficiency, in whom cellular immunity has been shown to be normal by all of the presently available tests.[69] The disease is extremely rare in otherwise healthy persons.[82]

The clinical association between pneumocystosis and starvation is solidly established and can be reproduced in the rat model mentioned previously (Figure 6).[83] Normal rats given either a regular or a low-protein diet continue to gain weight and show little evidence of pneumocystosis post mortem. In contrast, when rats are fed a protein-free diet, which produces weight loss and hypoalbuminemia, virtually all develop fatal pneumocystosis.

Treatment with corticosteroids has been a regular predisposing factor in most series of patients with P. carinii pneumonitis without prematurity, malnutrition or congenital immunodeficiency as an underlying factor.[69,72,79,80,84] This apparent selective ability of corticosteroids to induce P. carinii pneumonitis in patients is substantiated by the data of Frenkel and associates in the rat model, as he was able to induce P. carinii pneumonitis only with corticosteroids or cyclophosphamide (Cytoxan).[65] Total body irradiation, lymphoid ablation, methotrexate, chlorambucil, nitrogen mustard, 6-mercaptopurine and vinblastine did not induce pneumocystosis when used alone.

Recent studies by Masur and Jones[89] at Cornell may have defined a role for humoral antibody in resistance to P. carinii infection. When monolayer cultures of rat alveolar macrophages were exposed to Pneumocystis organisms obtained by lung lavage, the organisms attached to the outside of the macrophage cell envelope and remained there in a stable state for as long as several days. If antibody to Pneumocystis was added to the monolayer cultures, the attached parasites were rapidly phagocytosed and digested. Morphologically intact intracellular organisms were not observed, suggesting that P. carinii is not a facultative intracellular parasite. Antibody (with or without complement) added to Pneumocystis organisms in the absence of phagocytic cells had no apparent effect on the organisms; thus, antibody may act as an opsonizing agent essential to destruction of this organism by phagocytic cells. The role of other immune factors in resistance to Pneumocystis, such as thymus-dependent lymphocytes, cytotoxic lymphocytes or activated macrophages, remains to be defined. The recent development of the nude mouse as a model for P. carinii infection may allow further studies on the nature of immunity to Pneumocystis.[90]

TABLE 5.—*Conditions Predisposing to Pneumocystosis*

Prematurity
Starvation/malnutrition
Congenital immunodeficiency
Humoral
Cellular
Acquired immunodeficiency
Lymphoid malignancy
Drugs (especially corticosteroids)

Clinical Features

In the United States, the usual clinical features of a patient with P. carinii pneumonitis are those of an immunocompromised host who develops nonproductive cough, dyspnea (often with cyanosis as the disease progresses), respiratory distress, pulmonary infiltrates and hypoxia.[69,79,80,84-88] As mentioned previously, most patients with pneumocystosis have been receiving immunosuppressive drug therapy, particularly corticosteroids.[73,79,84,85,88] In the clinical studies that have

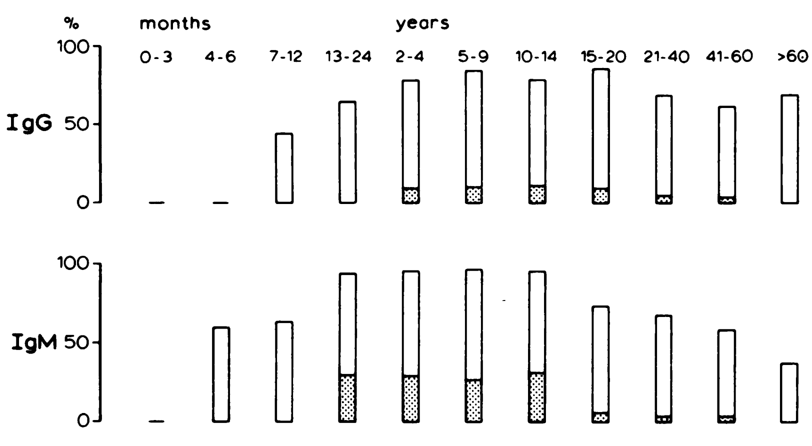

Figure 5.—Age distribution of IgG and IgM antibodies to Pneumocystis carinii in normal children, as determined by indirect immunofluorescence. Percentage of children with titers of 1:40 or higher (open bars) and 1:160 or higher (shaded bars) is shown. (Reprinted by permission from The University of Chicago Press and Meuwissen JHETh, Tauber I, Leeuwenberg ADEM, et al: J Infect Dis 136:43-45, Jul 1977.[74])

been reported, approximately 60 percent of the patients with corticosteroid-associated pneumocystosis have had their corticosteroid dosage reduced or discontinued shortly before the onset of significant pneumocystosis.[69,72,79,88] Virtually all patients have cough and dyspnea; hypoxia (arterial oxygen pressure of 80 mm of mercury or less while breathing room air) and hypocarbia also were present in most patients.[69,79,80,84,87] Hypercarbia may occur as the illness progresses. Pulmonary infiltrates tend to be symmetric and predominantly perihilar at first, with an interstitial and alveolar pattern. Later, extensive pulmonary consolidation with air bronchograms may be seen.[69,84-87] Atypical roentgenographic findings may occur; and patients with lobar consolidations, coin lesions and normal chest roentgenograms have been described.[91,92] The onset of illness is variable: it is more commonly acute with the patient progressing to severe respiratory failure over the space of two to ten days, but patients with a subacute or chronic evolution have been described.[84,86] Only half of the patients have fever, making this an unreliable sign of pneumocystosis.[84-87] The leukocyte count is not helpful, although lymphopenia or eosinophilia (or both) have been noted in some patients.[79] In the immunocompromised patient, death is usual if specific chemotherapy is not administered.

Although virtually all patients with pneumocystosis have disease confined to the lung, metastatic infection has been reported in a small number of cases.[93-96] When dissemination occurs, the organisms are found predominantly in regional lymph nodes (for example, perihilar nodes);[93] dissemination to more distant lymphoid tissues such as the spleen, liver and other lymph nodes is uncommon. Only rarely has P. carinii been found in tissues other than the lung and reticuloendothelial system.[94,96] Congenital infection can occur but is uncommon.[69] Multiple episodes of pneumocystosis have been described in individual patients; these probably represent relapses rather than recurrent infections.[85,97] Several instances of interstitial fibrosis occurring subsequent to P. carinii pneumonitis have been reported.[98] However, because interstitial fibrosis has occurred in patients with lymphoid malignancies and in patients receiving immunosuppressive therapy without P. carinii pneumonitis, it is premature to ascribe an etiologic relationship between Pneumocystis infection and subsequent interstitial fibrosis.

Dual Infections

Dual infections with cytomegalovirus were noted in the earliest clinical series of immunosuppressed patients with P. carinii pneumonitis.[73] The frequency of dual infections depends on the patient population studied (Table 6). In several series of patients with renal transplants, the incidence of coexisting cytomegalovirus infection in patients with pneumocystosis was quite high;[79,85] however, it has not been proved to be significantly higher than the incidence of cytomegalovirus infection in the transplant population overall. In other series of patients with tissue transplants or

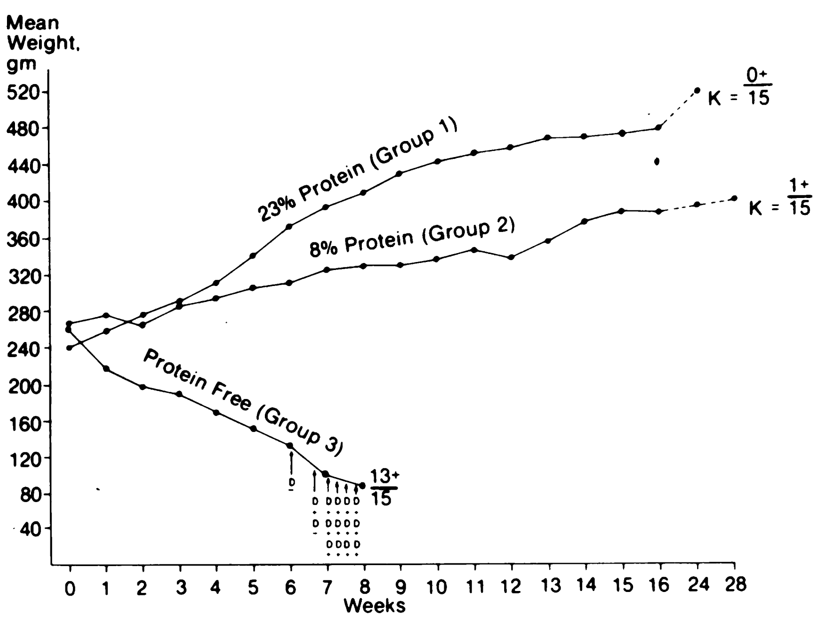

Figure 6.—Effects of various diets on body weights and incidence of Pneumocystosis in rats. Spontaneous death (D), killed (K), P, carinii found in lungs (+), P. carinii not found in lungs (−). (Reprinted by permission from Hughes WT, Price RA, Sisko F, et al: Am J Dis Child 128:44-52, Jul 1974;[83] copyright 1974, American Medical Association.)

TABLE 6.—*Clinical Association Between Pneumocystosis and Cytomegalovirus Infection*

Type of Patients	*Reference Number*	*Number of Patients*	*Percent Incidence of CMV in Patients with PC*
Renal transplants	114	7	71
Renal transplants	79	10	70
Transplants and malignancies	85	24	21
Transplants and malignancies	115	14	20
Malignancies	87	20	40
Malignancies	84	7	14
Malignancies (children)	80	51	10
Malignancies (children)	104	18	0

PC = pneumocystosis
CMV = cytomegalovirus

lymphoid and hematologic malignancies, the incidence of cytomegalovirus infection apparently was somewhat lower, although its presence was not always excluded rigorously.[80,87] A 33 percent incidence of antibody to P. carinii in immunocompromised patients with cytomegalovirus infection as reported by Norman and Kagan[75,76] suggests a relatively high frequency of dual infection overall. As will be discussed subsequently, there may be good explanations for why cytomegalovirus infection facilitates the subsequent development of Pneumocystis pneumonia.

Diagnosis

Making the diagnosis of P. carinii pneumonitis is part of the larger question of diagnosing the cause of pulmonary infiltrates in the immunocompromised host. The diagnosis of pneumocystosis rests on morphologic identification of the organism in tissues and secretions. This can be achieved using a variety of stains. Toluidine blue O, methylene blue and Gomori's methenamine silver stains have proved most satisfactory for demonstration of cysts and Giemsa stain for trophozoites.[69,99] Transtracheal aspirates reveal P. carinii in only 5 percent to 10 percent of patients with disease.[85,100] Bronchopulmonary lavage, either using a bronchoscope or a Carlen's catheter, has been used successfully to obtain P. carinii in some instances, although insufficient numbers of patients have been studied comparatively to determine the precise yield of this procedure.[101] Similarly, bronchial brushings have been successful in some instances, but usually the yield is not high and the material obtained contains artifacts that are difficult to distinguish from Pneumocystis cysts.[102] Needle aspiration of the lung has been used extensively and appears to produce an 80 percent to 90 percent yield with low complication rate.[80,85,100] Needle biopsy is an effective procedure for making the diagnosis, but because the rate of complications has been excessively high this procedure generally has been abandoned.[85] Therefore, when pulmonary tissue is required for histologic examination, either transbronchoscopic or open lung biopsy is utilized. Few studies have compared these two biopsy procedures in patients with pneumocystosis. However, bronchoscopic biopsy appears to be 80 perpercent or 90 percent as sensitive as open lung biopsy, although a recent report suggests it is even less sensitive.[103] Open lung biopsy is the diagnostic procedure by which other procedures are judged, but there are occasional reports of cases of pneumocystosis or other infections that have been missed even by open lung biopsy.[103]

TABLE 7.—*Open Lung Biopsy in Diagnosis of Pulmonary Infiltrates in the Compromised Host (47 Patients)**

Item	*Percent of Patients*	
Operative complications	0	
Pneumocystosis, total	35	
Alone		23
Other diseases	62	
Pneumonia (idiopathic)		29
Tumor		10
Fungal infection		10
Bacterial infection		8
Other		13

*Adapted from Rosen et al.[87]

Recently, investigators at Memorial Sloan-Kettering Cancer Center reported their experience with open lung biopsy in the diagnosis of pneumocystosis and other pulmonary infiltrates in the compromised host (Table 7).[104] Open lung biopsy studies were done in 47 patients with only a negligible rate of operative complications,[104] which has been the experience at other centers including the University of California, San Francisco. A third of the patients had P. carinii pneumonitis, and approximately two thirds of these had no other pulmonary abnormalities. Another 20 percent of the patients had either bacterial or fungal infections. Thus, over half of the patients had some sort of pulmonary infection, and it is likely that most of these would have been diagnosed by needle aspiration. On the other hand, nearly a third of the patients had idiopathic interstitial pneumonia without an associated infection, and an additional 23 percent had pulmonary tumor or other conditions, such as drug-induced pul-

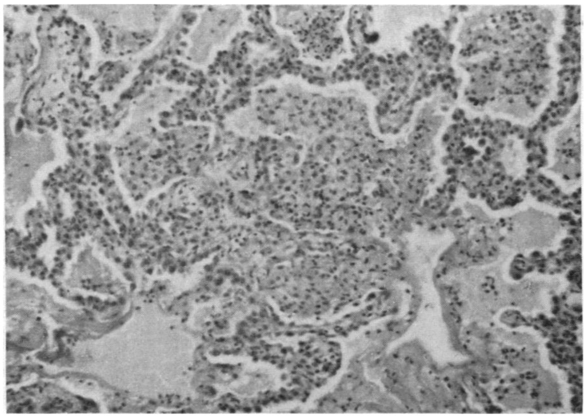

Figure 7.—Open lung biopsy specimen from a patient with pneumocystosis. Hematoxylin and eosin stain, ×400.

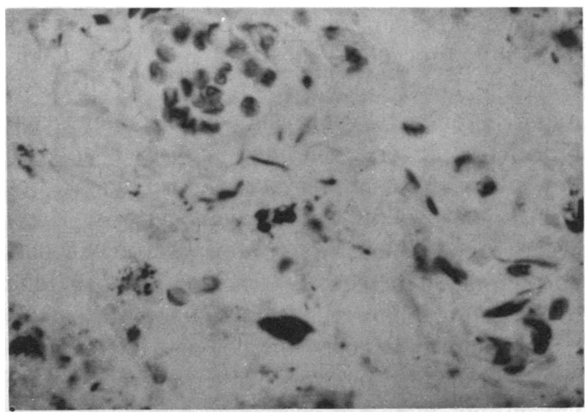

Figure 8.—Open lung biopsy specimen from a patient with pneumocystosis. Methenamine silver stain, ×800.

monary disease, that require histopathologic examination for specific diagnosis. Because effective therapy is available for some of the latter conditions, invasive diagnostic techniques are warranted in immunocompromised patients with undiagnosed pulmonary infiltrates. This is certainly true if needle aspiration is unrewarding or the infiltrate is progressing despite antimicrobial therapy.

With hematoxylin and eosin stains, the most prominent feature of early P. carinii pneumonitis is an eosinophilic foamy exudate filling the alveolar spaces, associated with a scanty mononuclear interstitial infiltrate.[69,105] This appearance is virtually pathognomonic of P. carinii pneumonitis. As the disease progresses, the interstitial infiltrate becomes dense and some fibrosis occurs (Figure 7). Silver stains show the alveoli filled with trophozoites (Figure 8). Touch preparations of lung biopsy material may show the cysts even more dramatically (Figure 9).

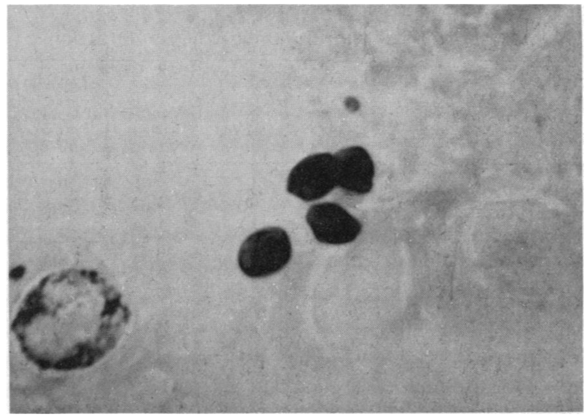

Figure 9.—Tissue touch preparation of open lung biopsy specimen from a patient with pneumocystosis. Methenamine silver stain, ×1,200.

TABLE 8.—*Pneumocystis carinii Antigenemia Detected by Counterimmunoelectrophoresis**

Patient Group	*Number of Patients*	*Percent with Antigenemia by CIE*
Documented pneumocystosis ...	123	68-95
Cancer, without pneumonia	100	15
Normal children	120	0

CIE = counterimmunoelectrophoresis

*Adapted from Pifer et al.[68]

There is some hope that serologic procedures will aid in the diagnosis of Pneumocystis infection. A complement fixation test for Pneumocystis antibody has been used in Europe for many years, particularly in infants and children. Because pneumocystosis tends to be more subacute in onset in these patients than in adult immunocompromised hosts, the test appears to be relatively effective and a detection rate of better than 75 percent has been reported.[57] Use of the complement fixation test in immunosuppressed adults has not been helpful.[76,79] A recently developed immunofluorescent antibody assay was tested by investigators at the Center for Disease Control in Atlanta, Georgia, and was positive in fewer than 1 percent of normal subjects and in fewer than 4 percent of contacts of patients with clinically apparent pneumocystosis; however, only 28 percent of immunosuppressed patients with documented P. carinii infection had antibodies.[75,76] Thus, the test is specific but relatively insensitive. A counterimmunoelectrophoretic assay for P. carinii antigen recently was developed by Pifer and associates,[77] and it appears considerably more promising for the diagnosis of acute disease (Table 8). In a series of 123 cancer patients with documented pneumocystosis, 68 percent to 95 percent had antigenemia. When fresh sera from 20 patients with documented pneumocystosis were tested with this assay, 19 were positive ($P < 0.05$). Of patients with cancer without pneumonitis, 15 percent were also positive, suggesting possible subclinical infection or reactivation. None of 120 normal children tested had detectable antigen. This test requires further evaluation but appears promising.

Treatment and Prophylaxis

Only a limited number of drugs are available for treatment of P. carinii infection. Pentamidine isethionate (Lomidine) and hydroxystilbamidine are effective in animals and until recently pentamidine was the standard drug used in humans.[88] Although effective in 50 percent to 70 percent of patients, it is quite toxic; and in the United States it is classified as an experimental drug available only through the Center for Disease Control. Recently, a controlled trial was conducted by Hughes and co-workers that compared trimethoprim-sulfamethoxazole (TMP-SMX) (Bactrim, Septra) with pentamidine in the treatment of P. carinii pneumonitis in children with acute lymphatic leukemia.[106] A preliminary study of TMP-SMX in cortisone-treated rats had shown that it was as effective as pentamidine in the treatment of P. carinii pneumonitis.[107] The success rates were comparable in the two patient treatment groups (77 percent compared with 75 percent), but the incidence of toxicity was significantly higher in the pentamidine-treated patients.[106] The dosage used was 20 mg of TMP and 100 mg of SMX per kilogram of body weight per day in four equal doses given by mouth. A parenteral preparation of TMP-SMX is under investigation, and this would facilitate therapy in extremely ill patients. Although the related drug combination of pyrimethamine-sulfadiazine is effective in patients with toxoplasmosis and has been shown to be curative in animal models of P. carinii pneumonitis, it has not received adequate clinical trials to allow determination of efficacy in patients.[108]

Based again on data from the rat model of P. carinii pneumonitis,[107] Hughes and his collaborators at St. Jude's Childrens Hospital in Memphis, Tennessee, undertook a controlled trial of TMP-SMX prophylaxis in children with acute lymphatic leukemia.[109] Pneumocystis pneumonitis did not develop in any of the 80 children treated prophylactically with TMP-SMX (5 mg of TMP and 20 mg of SMX per kilogram of body weight per day), whereas the illness occurred in 21 percent of the 81 children who received a placebo.[109] Consequently, all children with acute lymphatic leukemia at St. Jude's Childrens Hospital who are at high risk for pneumocystosis have received prophylactic treatment with TMP-SMX. Pneumocystis pneumonitis has virtually disappeared from this institution, despite an increase in the number of patients being treated for acute lymphatic leukemia (Figure 10). At present, TMP-SMX is the treatment of choice for P. carinii pneumonitis, and it should be used prophylactically in certain high-risk patients in whom the develop-

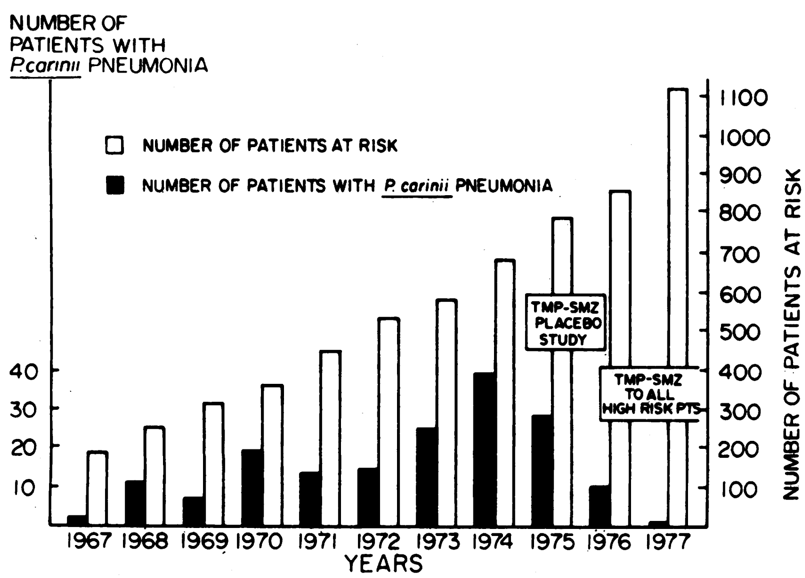

Figure 10.—Effect of trimethoprim-sulphamethoxazole (TMP-SMX) prophylaxis on the incidence of P. carinii pneumonia at St. Jude's Childrens Hospital, Memphis, Tennessee, 1967-1977. From October 1974 to October 1976, high-risk patients were randomly assigned to receive TMP-SMX or placebo; thereafter, all high-risk patients received TMP-SMX chemoprophylaxis. (Reprinted by permission from Hughes WT, Kuhn S, Chaudhary S, et al, and the *New England Journal of Medicine:* N Engl J Med 297:1419-1426, Dec 1977.[109])

ment of this condition is likely. Because instances of possible resistance of P. carinii to pentamidine have been reported, TMP-SMX prophylaxis should not be used indiscriminately.[69,88]

In summary, substantial advances have been made recently in our knowledge of P. carinii pneumonitis. The organism has been grown *in vitro,* and a possible role for antibody as an opsonin has been demonstrated in cell culture. Serologic studies have shown that the infection is prevalent in humans, suggesting reactivation as the major mechanism of clinical disease. Animal-to-animal aerosol transmission has been documented. Clinically, the relatively nontoxic drug TMP-SMX has been shown to be effective for both treatment and prophylaxis. A counterimmunoelectrophoretic assay for Pneumocystis antigen shows promise of being useful diagnostically. Sophisticated respiratory support has allowed survival of patients with respiratory failure through the period of antiparasitic drug therapy. However, there is still much need for investigation. The taxonomic position of the organism has not been determined precisely, and the antigenic and biologic relationships between animal and human strains have not been evaluated fully. The definitive reservoir (for human infection) has not been identified, and the spectrum of subclinical infection or disease has not been defined in either the normal or immunosuppressed host. Human disease is assumed to be due to reactivation of latent infection, but proof is lacking. Most important, we have no knowledge of how the organism produces disease, and the host defense mechanisms effective again P. carinii pneumonitis have not been defined.

Conclusion

From this discussion, it should be apparent that Pneumocystis and Toxoplasma share certain characteristics (Table 9). They are both found in man and in a wide variety of animals, usually in a latent form, emerging as opportunistic pathogens when host defenses are impaired. Toxoplasma is the more efficient pathogen of the two because it can cause disease in the normal host unlike Pneumocystis, which rarely, if ever, produces clinically apparent infection in a host with intact defense mechanisms. A large segment of the population has antibodies to Toxoplasma and Pneumocystis, reflecting widespread exposure. Both organisms are found in tissue as cysts and trophozoites. In the compromised host, both parasites exhibit tissue tropism, with Pneumocystis representing the purer example of organ predilection because it is rarely found in extrapulmonary sites. Toxoplasma has a predilection for the central nervous system, but it can affect virtually any organ. Neither organism can be cultivated on artificial media; they both require cell culture or laboratory animals for propagation. Toxoplasma is an obligate intracellular parasite, whereas the available evidence suggests that Pneumocystis exists primarily extracellularly. Effective therapy is available for both parasites, and it is remarkable that both drug combinations employed to treat infections with these organisms interfere with folic acid metabolism. Finally, both organisms are associated with concomitant cytomegalovirus infection in the setting of immunosuppression at a frequency that would suggest more than a chance relationship.

TABLE 9.—*Similarities Between Toxoplasma and Pneumocystis*

Factor	*Toxoplasma*	*Pneumocystis*
Host	Animal/human	Animal/human
Laboratory propagation		
Artificial media	−	−
Tissue culture	+	+
Laboratory animals	+	+
Infection normal host	Most asymptomatic	Most asymptomatic
Latency	+	+
Opportunistic pathogens	+	+
Intracellular parasitism	+	−
Tissue tropism	Brain	Lung
Treatment available	+	+
Concomitant infection by cytomegalovirus	+	+

In closing, let us consider some possible explanations for concomitant infections of Toxoplasma and Pneumocystis with cytomegalovirus. In experimental animals, infections with either cytomegalovirus or Toxoplasma can suppress cell-mediated immunity.[110,111] This segment of the immune system appears to be critical in the defense of the host against Toxoplasma[30] and cytomegalovirus[112] and likely is important in the control of Pneumocystis infection. Thus, the immunologic insult of a single infection, compounding others produced by treatment or malignancy, may well promote secondary infection. Another hypothetic explanation for concomitant parasite-virus infections might be an interaction of these organisms that would facilitate colonization of the potential host. For example, these two parasites might serve as a vector for cytomegalovirus. In fact, in a recent electron microscopic study of P.

carinii, structures were found within the cytoplasm of this parasite that resemble cytomegalovirus.[113] Finally, there may be some symbiotic relationship between these organisms in which elements essential to growth of one organism are provided by the other organism. Such a relationship has been suggested for Toxoplasma and cytomegalovirus because of the frequent finding in autopsy series of cells infected with both agents.[10] Lending support to this suggestion is a unique intracellular anatomic relationship of the two organisms that consists of Toxoplasma trophozoites surrounding inclusion bodies containing mature virus. One would not expect this relationship if dual infection of these cells was nothing more than a random event.

In summary, Toxoplasma and Pneumocystis are important opportunistic pathogens that account for significant morbidity and mortality in the compromised host. There is obviously a need to understand the pathogenic mechanisms responsible for disease production by these organisms. The occurrence of concomitant infection of these parasites with cytomegalovirus suggests more than a fortuitous relationship. If one could demonstrate a symbiotic relationship or an additive or synergistic suppressive effect of dual infection on the immune system, therapy directed at controlling one of these organisms might reduce the pathogenic potential of the other organism. Thus, an understanding of these relationships is more than of academic interest because it may suggest a means of controlling these important pathogens.

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